

Final Report

"Toxicology of Stranded Sea Turtles
as Related to the Stranding of July 10-13, 1990"

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I. Executive Summary

A stranding event comprised of 30 turtles occurred along the upper Texas Gulf coast during 10-13 July 1990. This event occurred 4 days after the Texas Closure ended on 6 July 1990 (beginning of the Texas Gulf shrimping season). Thirteen loggerheads (*Caretta caretta*) and 17 Kemp's ridleys (*Lepidochelys kempi*) stranded dead during this 4-day period. Necropsies were performed on all of these turtles. Liver, kidney and visceral fat were collected from 20 of the turtles and frozen for toxicological analysis. Prior to the 10-13 July strandings of 30 turtles, (March - June 1990), similar tissues were collected from 15 stranded turtles (5 loggerheads and 10 Kemp's ridleys). After the 10-13 July strandings, (25 July - 25 September 1990), tissues were collected from 20 stranded turtles (3 loggerheads and 17 Kemp's ridleys).

The objective of this evaluation was to attempt to explain the cause of death in the 10-13 July Stranding event turtles. Out of the 55 sea turtles from which samples were collected, liver and kidney from 10 loggerheads were randomly selected for heavy metal analysis by Atomic Absorption Spectroscopy. Pathological data are shown in Table 1 for these 10 loggerheads (six were from the 10-13 July's stranding event and four controls consisted of two pre-July 10 and two post-July 13 turtles).

Selected heavy metals were measured to provide comment as to the health risk posed by these environmental pollutants. Zinc (Zn), mercury (Hg), iron (Fe), arsenic (As), cadmium (Cd), selenium (Se), lead (Pb), and copper (Cu) were analyzed (Tables 2 and 3). Based on results for terrestrial animals (Table 4) and

fresh-water and sea turtles (Table 5), none of the metal levels, except for perhaps As and Cd, were high. The Cd levels in these turtles are similar to levels we have measured in bowhead whales from 1984 to 1990. The kidney Cd levels in this study were higher than levels reported in sea turtle eggs or in freshwater turtle kidneys (Table 5). However, they did fall within a range reported for several marine mammals (Tables 9 and 10).

The As levels in the liver and kidney of stranded turtles were higher than levels we found in bowhead whales (Tables 9 and 10), and would be in the toxic range in cattle according to Puls (1988). These As levels would not be in the toxic range in the dog, horse, or pig but would be at the higher end of normal according to Puls (1988). There are data to show that in marine fish, other sea animals, and from our work in one whale, the As is about 90% in the form of arseno-betaine which is not very toxic (Cannon *et al.*, 1983; Vahter *et al.*, 1983). This could be true in turtles, but this analysis is very time consuming and expensive.

Zinc and Hg levels in stranded turtle tissues were higher than those in the bowhead whale (Tables 9 and 10), but toward the lower levels reported for most whales and dolphins. Fluctuating Fe levels in these tissues were probably due to the varying amount of blood present in the tissues when collected. A statistical comparison of liver and kidney metal concentrations of the four loggerhead turtles collected outside the 10-13 July period to those of six conspecifics found within the 10-13 July stranding period indicated no significant differences between the two time periods. Overall, turtles stranding on 10-13 July had slightly lower levels than did turtles stranding prior to or after 10-13 July.

Inability to detect differences in metal levels between the stranding groups is partly due to the large variability in the data. Using these data as a pilot study, there is some evidence that increasing the sample size might allow for detection of differences between the groups for some metals (additional observations on arsenic might be helpful).

Additional observations are needed to accurately compare metal levels in the two species (Kemp's ridley and loggerhead). Moreover, samples from sea turtles which have not been exposed to the pollution sources in Galveston Bay or the Gulf of Mexico are needed to define "normal" metal levels. These tissues could be obtained from turtles that die in captivity. The number of additional turtles needed to provide quality results is not clear. We recommend that the second phase of this project be initiated by performing a heavy

metal analysis with liver and kidney tissues from 30 turtles (10 loggerheads and 20 Kemp's ridleys) which are in storage.

Concentrations of 15 organochlorine pesticides and 5 polyaromatic hydrocarbons (PAHs) were determined from visceral fat samples (Table 7). No consistent pattern of organochlorine pesticides was found in the 10 turtles (seven loggerheads and three Kemp's ridleys) tested (Table 8). The amounts that were present in five of the turtles were insignificant. Deteriorated condition of tissue samples may have contributed to this sporadic identification of toxic residue. No PAHs were detected in any tissue sample. The absence of PAHs and low levels of pesticides suggests these pollutants did not pose a health risk to the sea turtles during the period July 10-13.

Analyses of loggerhead kidney and liver samples indicated Zn, Fe, Se, Pb, and Cu levels were within ranges reported for freshwater turtles (Table 5), sea turtles (Table 5) and marine mammals (Tables 9 and 10) and probably would not be detrimental to the health of sea turtles. Mercury levels were within the range reported for other animals (Tables 5, 9 and 10). The As levels would not appear toxic but on the high normal side in comparison to those for the dog, horse, and pig (Puls, 1988). The Cd levels were high in comparison to those reported by other turtle studies (Table 5); however, they were within the range reported for whales and dolphins (Tables 9 and 10) and generally below levels reported for terrestrial animals (Puls, 1988).

II. Introduction

Enhanced concern for the integrity of the coastal environment in the Gulf of Mexico has generated interest in the phenomenon of beach strandings as it relates to highly visible species such as sea turtles. Increased documentation of sea turtle strandings on Texas beaches in recent years has led to questions regarding the causes of apparently elevated mortalities. Texas is the site of three major industries - chemical, shrimp and oil - whose operations are perceived in conflict with sea turtle stocks.

Marine pollutants are attracting increased attention as environmental awareness continues to grow. Several events, including recent major oil spills, a large sea turtle stranding event on 10-13 July 1990, and a growing number of environmentally concerned citizens, have created an interest in how pollutants are affecting the environment and sea turtles.

Many chemical pollutants, introduced anthropogenically or naturally occurring, could possibly affect sea turtles and other marine animals. Petroleum hydrocarbons (oil and its derivatives) are continuously released in large volume by natural seepage from the sea floor. But it is the single release of large amounts of oil, such as the release of oil in Kuwait by Iraq, the Exxon Valdez spill, or the Megaborg spill, that generates the most press coverage. Organochlorines, such as DDT and its more toxic decomposition compounds, DDD and DDE, are anthropogenic pesticides which have entered and remain in the marine environment. Like petroleum, heavy metals are naturally occurring substances, used extensively in industry and even in the household. They can occur locally in unusually high and potentially harmful concentrations. Certain heavy metals, such as Cu, Se, Fe, and Zn, are essential for proper metabolism, but even these elements can be toxic at high concentrations.

The effects of pollutants on sea turtles are difficult to assess because the animals are not confined to an easily studied habitat. The size and life span of turtles as well as regulatory restrictions on endangered and threatened animals prevent contained laboratory studies of these animals. This paper focuses on measurement of heavy metal concentrations, organochlorine pesticides, and polyaromatic hydrocarbons found in sea turtles stranded on the Texas coast.

A large stranding event comprised of 30 turtles occurred along the upper Texas Gulf coast during 10-13 July 1990. This event occurred four days after the Texas Closure ended on 6 July 1990, marking the reopening of the Texas Gulf shrimping season. Thirteen loggerhead, *Caretta caretta*, and 17 Kemp's ridley, *Lepidochelys kempi*, sea turtles stranded during this 4-day period. Necropsies were performed on all of these turtles. Liver, kidney and visceral fat were collected from 20 of the turtles and frozen for toxicological analysis. Similar tissues were collected from 15 stranding turtles (10 ridleys and 5 loggerheads) prior to 10-13 July (March - June 1990). Tissues also were collected from 20 turtles (17 ridleys and 3 loggerheads), stranding after the 10-13 July (July - September 1990).

The National Marine Fisheries Service, in a letter dated 29 January 1991, stated their desire to fund a toxicological analysis of these tissues in two phases. The first phase would include tissue analysis of six turtles from the 10-13 July strandings, two pre-July 10 and two post-July 13 turtles. The second analytical phase would include 20 turtles, ten from the 10-13 July stranding and five pre-July 10 and five post-July 13 turtles.

This report pertains to tissue analyses in the first phase of the project.

Toxicology

Drs. Raymond Sis and Andre Landry have been involved in a MARFIN sea turtle research project, "Assessment of Nonshrimping Mortality of Sea Turtles," from which tissues were obtained for the analyses discussed in this report. Toxicological information from stranding/necropsy data collected by NMFS, STSSN and TAMU may assist in characterizing causes of sea turtle mortality. This MARFIN study may permit an assessment of natural and human related factors in the mortality patterns of these species in the U.S. Gulf of Mexico coastal waters. Such research is critical to survival of endangered sea turtle stocks, as it generates data on the role toxins play in sea turtle deaths. Future survival of sea turtle stocks in the Gulf of Mexico will depend on such research findings.

III. Purpose

A. Description of Problem

Very little baseline information is available concerning the levels of environmental contaminants which occur in marine animal tissues. It is imperative that levels of various relevant chemical pollutants within the tissues of key marine animals be defined. This baseline information will be necessary if realistic comparative assessments are to be made.

We selected 15 organochlorine pesticides and 5 polyaromatic hydrocarbons (PAHs) (Table 7) for this study because these were the only ones that were found in previous work in whales. We selected visceral fat for the analysis because organochlorines and PAHs are lipid soluble and, therefore, sequestered in fat. Organochlorine pesticides and PAHs have greater affinity for tissues high in lipid content (Meeks, 1968; Pearson *et al.*, 1973; Stone *et al.*, 1980; Helwig and Hora, 1983; Watson *et al.*, 1985; Bryan *et al.*, 1987). We selected eight heavy metals because: 1) these are the ones likely to be a problem on the Texas Coast; 2) they are the same metals selected for our previous tissue analysis work in whales; and 3) they have been the most commonly studied in previous marine animal studies. Copper (Cu), iron (Fe), zinc (Zn), and selenium (Se) are essential trace metals that can reach toxic levels. Arsenic (As), lead (Pb), mercury (Hg), and cadmium (Cd) are non-essential trace metals that

can be potentially toxic at certain levels.

Non-essential trace metals such as Cd and Hg accumulate with age in marine mammals, increasing by over 200 fold from fetuses and juveniles to larger adults. Itano *et al.* (1984) determined that mercury in striped dolphin liver of fetuses is 100 fold lower than in mature female striped dolphins. A dramatic increase with age has also been observed in livers and other tissues of Texas stranded bottlenose dolphins (Arima and Nagakura, 1979).

The distribution of trace metals in dolphins stranded in Texas seems to be tissue dependent (Tables 9 and 10). Liver and kidney were the tissues selected for this study because they have been found to have higher concentrations of metals than did muscle or fat (Hammond and Beliles, 1979). We found that some heavy metals concentrate more highly in the liver, while other heavy metals concentrate more highly in the kidney. In many cases the liver is the greatest concentrator of metals (i.e., Hg and As). Bone and shell were found to contain the highest concentrations of Pb, followed by liver and kidney tissues of freshwater turtles (Beresford *et al.*, 1981; Krajicek and Overman, 1988). However, the largest quantities of Cd are generally found in kidney in turtles (Robinson and Wills, 1975) and in porpoises (Fujise *et al.*, 1988).

B. OBJECTIVES

1. To determine the levels of certain heavy metals, organochlorine pesticides, and polyaromatic hydrocarbons (PAHs) in selected tissues of stranded sea turtles.
2. To provide comment as to the health risk posed by these environmental pollutants to the animals themselves by comparing the levels of toxins found in turtle tissues with those found in other marine animals and selected terrestrial animals.

IV. Approach

A. Detailed Description of the Work

Toxicological analysis was performed on tissues from 10 sea turtles (six were from the 10-13 July stranding event and four were controls, two pre-July 10 and two post-July 13) in the following manner:

Heavy Metals: Samples of liver and kidney from 10 loggerheads were collected during necropsies. A preliminary analysis showed a species difference in levels of heavy metals between loggerheads and Kemp's ridleys. This necessitated a conspecific analysis (Tables 1-3). Loggerheads were chosen for the heavy metal analysis because they are the most abundant species in the Gulf of Mexico. The tissues were excised with a stainless steel knife, placed in individual polyethylene bags, frozen, and transported in a styrofoam container to the TAMU College of Veterinary Medicine.

Laboratory processing: Sample processing methods were designed to minimize metal contamination. Samples were slightly thawed to facilitate cutting, and a stainless steel scalpel was used to trim the edges of each tissue to eliminate any contamination that may have been introduced during field collection and necropsy. Approximately 1 g each, of the liver and kidney samples was placed into a Savillex teflon digestion bomb and digested with 3 ml of Ultrex ultrapure nitric acid for 8 hours at 120°C. This procedure insured that all organic matter was oxidized to carbon dioxide and that all metals were brought into solution. The sample digest was diluted with 20 ml of deionized distilled water and transferred to a polyethylene screw-cap bottle for storage. Blank samples and National Research Council Canada Dolt-1 and Dorm-1 (reference material with known metal concentrations) were digested and analyzed simultaneously to assure quality control. Values for these reference materials are reported in Appendix A.

Mercury concentration was measured by cold-vapor atomic absorption spectrophotometry (AAS), Zn and Fe by flame AAS and the remaining elements by graphite furnace AAS using techniques developed during the NOAA National Status and Trends Program. Samples from five loggerheads were analyzed by neutron activation analysis to confirm AAS accuracy (Table 2). This gave us the opportunity to look at the concentrations in the tissues, with two methodologies. The neutron activation data helped confirm the fact that we had accurate data.

Tissue samples were reported in µg metal/g of tissue on a wet weight basis for all metals. Tissue moisture was recorded so that conversion to µg/g dry weight could be accurately done in order to compare to other published metal investigations if necessary.

Determination of tissue levels of organochlorines and polyaromatic hydrocarbons (PAHs):

Samples of visceral fat from 10 sea turtles were collected during necropsies. The 10 visceral fat tissues were collected and processed from the same turtles that the liver and kidney samples were collected. The fat tissues were submitted to the Texas Veterinary Medical Diagnostic Laboratory for organochlorine and PAH analysis. Table 7 represents the 15 organochlorine pesticide standards and 5 PAH standards that were analyzed. A preliminary analysis with both loggerheads and Kemp's ridleys showed no difference between species in the organochlorine and PAH determinations; therefore, data in this portion of the study is based on both loggerheads and Kemp's ridleys (Tables 6 and 8).

Laboratory Tissue Preparation: Frozen samples of sea turtle visceral fat (minimum wet weight of 2 g) were trimmed to avoid field contamination, cut from larger pieces, and weighed on a top loading balance to ± 0.01 g for extraction preparation. Visceral fat samples from seven loggerheads and three Kemp's ridleys were analyzed for organochlorines and PAHs.

The samples were minced with scissors. Ten ml of a 50:50 mixture of dichloromethane-cyclohexane was added to the sample, and the mixture was finely ground by a tissue grinder (Tissumizer TM). Dichloromethane-cyclohexane was used to rinse all materials (scissors, etc.) required during sample preparation to insure minimal lipid contamination. The ground samples were placed in pre-weighed screw-top tubes which were then weighed. The uncapped tubes were placed under a hood, and the organic mixture was allowed to dry to a constant weight. Lipid concentrations were calculated by difference in weight of dried organic mixture from ground samples.

The dried fat samples were thoroughly mixed with 3 g of Bondesil TM C18 using a glass mortar and pestle. This mixture was loaded to the top of a 20 cc plastic syringe that was filled with a bottom layer of 10 cc of florosil and a top layer of 5 cc Bondesil TM C18 which was previously washed with HPLC grade acetonitrile. Organochlorines and polyaromatic hydrocarbons were eluted from the columns with 15 ml of HPLC grade acetonitrile. Remaining acetonitrile was forced off the column with air using a rubber bulb. The acetonitrile was evaporated under a high purity nitrogen stream in a water bath at 65°C from the eluted mixture, and the residue was resuspended in 50 μ l of methanol.

Dilutions were prepared from these stock resuspensions. To determine accuracy of recovery the resuspended samples were also spiked with aldrin.

Instrumentation: Resuspended, diluted pesticide standards and samples were determined by a Shimadzu Gas Chromatograph/Mini 2 with an electron-capture detector. The column used was a 30 m long (0.25 μ m film thickness and a 0.25 μ m internal diameter) Supelco SPB-5 column which was packed with 5% diphenyl, 94% dimethyl, and 1% vinyl polysiloxane.

Temperature during the analytical process was controlled as follows: with the injection port at 250°C, initial temperature of 165°C was held for 3 minutes and then raised to 280°C at a rate of 5°C/minute. The final temperature was held for 3 minutes.

Resuspended and diluted PAHs and standards were analyzed with a Hewlett Packard 5790A Gas Chromatograph with a mass spectrometer detector using a Hewlett-Packard Ultra 1 column cross-linked with methyl silicon. The column was 25 m long and 0.2 mm inner diameter with a 0.33 μ m film thickness.

The injection port was set at 250°C with an initial temperature of 60°C held for 1 minute. A temperature program of 15°C/minute was used until a final temperature of 280°C was reached. Final temperature was held for 23 minutes.

Peak areas from the samples were determined and compared to peak areas from known reference standards. Total nanograms (ng) of each pesticide and PAHs were calculated and then converted to ng/g (ppg) lipid weight and ng/g (ppg) wet tissue weight.

V. Findings

A. Work Accomplishments and Findings

During the period 10-13 July 1990, 30 sea turtles stranded along the upper Texas coast. The turtles were brought to the National Marine Fisheries Service (NMFS) Galveston Laboratory for necropsy by Texas A&M University (TAMU) in an effort to determine cause of death and gather other life history information. These included 17 Kemp's ridleys and 13 loggerheads. Eight of the Kemp's ridleys were females and nine were of unknown sex. One female Kemp's ridley possessed an external

flipper tag indicating it was a head started turtle.

Table 1 is a summary of the necropsy data for the 10 stranded loggerheads (six from the 10-13 July strandings, two pre-July 10 and two post-July 13 turtles) used for the heavy metal analysis in this project. Table 6 is a summary of the necropsy data from the seven loggerheads and the three Kemp's ridleys used for the organochlorines and PAH analysis.

Necropsies conducted by a TAMU veterinarian did not elucidate the cause of death for the 30 turtles stranding 10-13 July. One Kemp's ridley turtle suffered a shark attack either pre- or postmortem. Three turtles appeared to have undergone human-induced mutilation: one Kemp's ridley was missing the soft tissue around its rear flippers, tail and neck; a second ridley was missing the head, front flippers and left rear flipper; and a loggerhead was missing rear flippers. All three turtles possessed straight-edged cuts along these areas suggesting they were mutilated rather than cut by scavengers such as sharks. Two of these turtles were found in a fresh condition by a private citizen, who indicated the front flippers on the loggerhead had rips as if it had been hooked with a gaff and then the hook torn out of the flesh. He also noticed blood coming from the turtle's mouth and scuff marks and breaks in the skin on the top of the head between the eyes. A second Kemp's ridley also had worn edges on the shell. It had a puncture between its eyes, the wound being about 2.5 cm long and 3 mm wide and apparently caused by a knife. Both turtles were relatively fresh because blood was still flowing from the wounds. It appears that these two turtles were deliberately mutilated.

Preliminary analysis of the gastrointestinal contents indicate that the Kemp's ridleys were feeding on assorted crabs (*Callinectes*, *Persephona*, *Libinia*, *Menippe*, *Hepatus*), as well as some small portunid crabs, shrimp and *Leptogorgia* (gorgonian). Loggerheads fed on *Diopatra* (tube worm), assorted fish, anemones, shrimp and crabs (*Callinectes* and *Persephona*). No oil or plastics were found in the intestinal tracts.

We statistically analyzed data on heavy metal concentrations in the liver and kidney of the 10 loggerhead turtles. Specifically, for each organ, we compared seven metal concentrations (Zn, Hg, As, Cd, Se, Pb and Cu) of the four loggerheads collected outside the 10-13 July period to concentrations

in the six loggerheads found during the 10-13 July stranding period. We found no statistically significant differences between heavy metal levels of the two time periods. Some of the differences tended to indicate that the 10-13 July stranded loggerheads had lower levels than the control loggerheads.

Based on data for other animals, none of the metals, except for perhaps As and Cd, were high in this study (Tables 2 and 3). Puls (1988) reviewed literature regarding metal toxicity in domestic animals and published tables establishing so-called normal levels of metals in tissues from cows, dogs, horses, and pigs (Table 4). He also reported so-called "toxic" levels of metals in organs from animals that had been poisoned. Cadmium levels in the loggerheads were similar to levels we have measured in bowhead whales (Tables 9 and 10). The whales appeared to be normal. Cadmium typically accumulates in liver and kidney rather than in muscle and has a long half-life in biota (Dycus, 1986). Low levels of Cd, 0.01 $\mu\text{g/g}$, were found in some snapping turtle muscle tissue samples (Helwig and Hora, 1983), while Dycus (1986) detected an average concentration of 0.33 $\mu\text{g/g}$ in liver samples and 0.28 $\mu\text{g/g}$ in snapping turtle muscle samples. He stated that Cd found in muscle may have been related to low level accumulation over a long period of time because turtles are long lived and Cd is persistent. Dycus noted that the presence of Cd in snapping turtle livers indicates a need for further evaluation because this metal is highly toxic to aquatic life. The kidney Cd levels in loggerhead turtles measured in this study ($18.88 \pm 14.83 \mu\text{g/g}$) were higher than those reported in loggerhead eggs (Stoneburner *et al.*, 1980; Hillestad *et al.*, 1974), in snapping turtle kidneys (Albers *et al.*, 1986) and in softshell turtle kidneys (Robinson and Wells, 1975) (Table 5). However, our readings fell within a range reported in several species of whales and dolphins (Table 9).

The As levels in the liver and kidney of the 10 stranded loggerheads were higher than levels found in bowhead whales (Tables 9 and 10), and within the toxic range in cattle according to Puls (1988). The As levels in loggerheads were not in the toxic range, but on the high normal side in the dog, horse, and pig according to Puls (1988). We did not find any reports for As levels in any turtle species. There are data to show that in marine fish, other sea animals, and from our work in one whale

the As is about 90% arseno-betaine which is not very toxic (Cannon *et al.*, 1983; Vahter *et al.*, 1983). This could be true in sea turtles, but this analysis is very time consuming and expensive.

Mercury levels were higher than levels we found in the bowhead whale (Tables 9 and 10). Mercury levels reported in snapping turtles (Table 5) were 0.2 - 0.4 µg/g (Bertrand, 1974), 0.5 µg/g (Garcia and Kidd, 1972), 0.02 - 0.21 µg/g (Helwig and Hora, 1983), 0.39 - 1.28 µg/g in liver and kidney tissues (Albers *et al.*, 1986), 0.27 µg/g in liver tissue (Dycus and Lowery, 1986), 0.12 - 1.3 in kidney tissue (Meyers-Schone and Walton, 1990) and 0.33 µg/g in liver tissue (Dycus, 1986). The Hg concentrations in kidney and liver tissue in our study fell within the ranges reported above, with the exception of one pre-July 10 loggerhead (90051102) with a reading of 2.46 µg/g in the liver. This level is just in the range considered toxic in cattle (Table 4) but within the reported range in marine mammals (Table 10). The Japanese consider the maximum allowable limit of Hg in fish products for human consumption to be 0.4 µg/g.

Levels of Zn, Fe, Pb, Se, and Cu levels were similar to and within the range of levels reported in terrestrial (Table 4) and marine animals (Tables 9 and 10) and fresh-water turtles (Table 5). Our concentrations were similar to those found in an analysis on snapping turtle fat and livers which indicated levels of metals (Cu, Fe, Se, Zn) were generally low and probably not impacting Gunter's Reservoir biota (Dycus and Lowery, 1986).

Very little is known about baseline levels and physiological effects of heavy metals in sea turtles. Hall (1980) recorded some environmental contaminants in reptiles. Witkowski and Frazier (1982) published on heavy metals in cheloniid turtles. They ran a pilot study using bone and barnacle samples. Zinc (46 to 955 µg/g) and iron (78 to 309 µg/g) levels in the bone and barnacle were greater than copper (8.6 to 18 µg/g), manganese (8.4 to 35.6 µg/g), and lead (41.5 to 97.2 µg/g) levels. This was consistent with findings of Stoneburner *et al.* (1980) who worked with egg yolks in loggerheads. Stoneburner's levels of Zn and Pb in loggerhead eggs were higher than the current findings in loggerhead kidney and liver. Our levels of Fe and Cd in loggerhead tissues were higher than their findings in the loggerhead eggs. The comparisons indicated that eggs laid on different beaches had

significantly different mean concentrations of Ba, Co, Cr, Hg, Mo, Ni, and Pb.

Loggerhead eggs from three nesting beaches in Georgia and South Carolina were analyzed for heavy metals (Hillestad *et al.*, 1974). Mercury levels in yolks ranged from 0.02 ppm to 0.09 ppm and 0.01 ppm to 0.03 ppm in the albumen. Zinc levels averaged 32.25 ppm in yolk and 26 ppm in the albumen. There were significant differences between the yolk and albumen levels of Cd, Cu, and Pb. Cadmium levels averaged 0.17 ppm in yolk and 0.56 ppm in albumen; Cu levels averaged 2.08 ppm in yolk and 6.0 ppm in albumen; Pb averaged 2.87 ppm in yolk and 12.0 ppm in albumen.

The inability to detect statistical differences in metals between the loggerhead stranding groups in our study is partly due to the large variability in the data. Using these data as a pilot study, there is some chance that increasing the sample size might allow detection of differences between the groups for some metals (As is one metal where additional observations might be helpful). It would also be desirable to obtain samples from loggerheads which have not been exposed to the pollution sources in Galveston Bay and Gulf of Mexico in order to attempt to define "normal" levels of metals. These samples could be obtained from sea turtle carcasses that become available from deaths of turtles held in captivity, deformed turtles which are sacrificed for scientific purposes, or from accidental deaths. It also would be meaningful to compare loggerheads and Kemp's ridleys. The number of additional turtles needed to provide more definitive results is not clear. We have tissues in our freezer from 27 turtles (4 loggerhead and 10 Kemp's ridley controls, and 3 loggerhead and 10 Kemp's ridley stranding event). We recommend that the second phase of this project be initiated by performing a heavy metal analysis on the remaining turtle tissue in our freezer, plus perform analyses on turtles that die naturally (unexposed to the coastal environment) or non-exposed controls. Meyers-Schone and Walton (1990) indicated that variations in diet between species play a significant role in the exposure of snapping and yellow-bellied turtles to Hg. Completing the second phase of the study using loggerheads and Kemp's ridleys would provide a combined baseline data set for 20 loggerheads and 23 Kemp's ridleys. From this, we could then publish some kidney and liver data in the literature which other investigators might find worthwhile for future monitoring studies in the Gulf of Mexico. In addition to the above 40

turtles, we have tissues collected and stored from 14 Kemp's ridley and 10 loggerhead turtles which stranded between October 1990 and April 1991. The heavy metal analysis of these 24 turtles would add to the data base and make the data more statistically significant.

The fact that most all turtles are both long-lived and mobile allows for the integration of exposure over time and space (Meyers-Schone and Walton, 1990). In addition, liver, kidney, fat, muscle and bone, key tissues useful in toxicological analyses, are available in large quantities in stranded sea turtles.

Determination of tissue levels of 15 organochlorine pesticides and 5 polyaromatic hydrocarbons (PAHs) were made from visceral fat taken from seven loggerheads and three Kemp's ridleys (Table 7). There was no pattern of organochlorine pesticides present in the 10 turtles tested (Table 8). The amounts that were present in five of the turtles were insignificant. The deteriorated condition of the tissue may have contributed to this sporadic identification of the toxic residue. There is controversy whether pesticides are diluted over time in post-mortem animals. A fresh sample may be needed for good results. The pesticides may leach out during decomposition. Baseline analyses are needed on fresh sea turtle tissues.

In a recent study of post-mortem changes in freshwater red-eared slider turtles (*Pseudemys scripta elegans*), we found that after death these turtles first sink to the bottom and remain there for 1 1/2 to 2 days. After the body cavity fills with gas, the carcass floats to the surface. The dead turtle will be 2 to 3 days post-mortem before it will be carried by the tides, waves, and winds to the beach. By this time the tissues begin to soften and autolysis has started. Serosanguinous fluid from the tissues is found in the body cavities. Therefore, this suggests that dead stranded sea turtles are not a good source of tissues for toxicologic analysis of some pesticides. The preferred tissues would come from freshly dead sea turtles.

Since autolysis of the liver, kidney and visceral fat take place after 8 hours post-mortem, it is difficult to obtain a satisfactory tissue sample for toxicologic analysis. It may be that fresh tissues are needed to obtain consistent results with organochlorine analyses. The presence of DDT and its

metabolites has been the most frequently reported pesticide in the freshwater snapping turtles (Meeks, 1968; Flickinger and King, 1972; Reeves *et al.*, 1977; Punzo *et al.*, 1979; Stone *et al.*, 1980; Albers *et al.*, 1986), loggerhead and green sea turtles (McKim and Johnson *et al.*, 1983), and in the eggs of loggerhead turtles (Hillestad *et al.*, 1974; Clark and Krynitsky, 1980, 1985). Clark and Krynitsky (1980) found that DDE and PCBs (in loggerhead eggs) declined significantly in a 10-day interval during incubation. They reported on a second study with loggerhead eggs (Clark and Krynitsky, 1985) in which they detected amounts far below levels thought to be harmful. Samples of ten green turtle eggs were obtained for PCB and pesticide analysis (Thompson *et al.*, 1974). Levels of 0.001 to 2.52 ppm were determined. In loggerhead sea turtle eggs from nesting beaches in Georgia and South Carolina, Hillestad *et al.* (1974) reported total DDT (DDE + DDD + DDT) residue levels ranged from .058 ppm to .305 ppm. Dieldrin residue levels ranged from trace to .0564 ppm.

No amounts of PAHs were detected in the loggerhead and Kemp's ridley tissue in this study. Tissue analysis in heavily oiled sea otters has not been dramatic (Davis, 1991). Petroleum hydrocarbons may be rapidly cleared. Blood petroleum hydrocarbons measured when heavily oiled sea otters first came in during the Valdez oil spill had some high levels. However, levels from tissues taken during necropsy did not identify any elevations. It may be difficult to find PAHs in tissues. Because no PAHs were detected and the amounts of pesticides detected were insignificant in the tissues from the dead stranded loggerhead sea turtles, we recommend that we do not initiate the analysis of PAHs and pesticides in the proposed phase 2 of this study.

Low levels of pesticides and the absence of PAHs indicated these environmental pollutants do not pose a health risk to loggerhead and Kemp's ridley sea turtles. Analyses on loggerhead kidney and liver indicated Zn, Fe, Se, Pb, and Cu levels were within ranges reported in freshwater turtles, sea turtles, and marine mammals and probably did not detrimentally impact the health of the loggerheads. Mercury levels were within the range reported in other animals with the exception of one level in a loggerhead stranded prior to the stranding event. The As levels would not appear toxic but on the high normal side in comparison to the dog, horse, and pig. The Cd levels were high in comparison to other

turtle findings; however, they were within the range reported in marine mammals and not toxic within the range reported for some terrestrial animals (Puls, 1988).

Using these data as a pilot study, there is a chance that increasing the sample size might allow detection of differences between the groups for some metals. Additional observations also are needed to be able to accurately compare the two species (Kemp's ridley and loggerhead). It would be helpful to obtain samples from sea turtles which have not been exposed to the pollution sources in Galveston Bay and the Gulf of Mexico in order to attempt to define "normal" levels of metals. We recommend that the second phase of this project be initiated by performing a heavy metal analysis with tissues from 27 control and stranding event sea turtles which are in storage.

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TABLE 1. Summary of necropsy data for 10 loggerhead sea turtles utilized in heavy metal analyses related to the stranding event of 10-13 July 1990.

| ID AND TIME PERIOD | SPECIES | SCL(cm) STRAIGHT CARAPACE LENGTH (cm) | CONDITION CODE** | EXTERNAL ABNORMALITIES | COUNTY | SEX | G.I. CONTENTS COLLECTED/ MAJOR ITEMS | TOXICOLOGICAL SAMPLES | MISCELLANEOUS * COMMENTS |
|----------------------------|---------|--|---------------------|--|-----------|-----|--|--|---|
| 90030801 (Pre-July 10) | CC* | 60.6 | 2 | Head gone; carapace disarticulating | Matagorda | F | Yes/crabs | Liver, kidney, visceral fat | Cuts near head, apparently straight edge |
| 90051102 (Pre-July 10) | CC | 67.5 | 2 | Head & all flippers gone | Galveston | F | Yes/unknown | Liver, kidney, visceral fat | Straight edge cuts on carapace |
| 90071001 (July 10-13) | CC | 57.2 | 2 | Black material (tar) collected from plastron & Lt. rear flipper; epizoa collected | Galveston | M | Yes/ crabs, worms & anemones | Liver, kidney, visceral fat, blood | Dorsal tip of Rt. front flipper exhibits healed wound |
| 90071002 (July 10-13) | CC | 76.5 | 2 | Skin sloughing on head; epizoa collected | Galveston | F | Yes/worms | Liver, kidney, visceral fat, blood | Fish hook & fishing line found in tissue dorsal to esophagus; right atrium of heart exhibits air bubble |
| 9071101 (July 10-13) | CC | 56.8 | 2 | Scutes sloughing on shell & flippers; left marginal bones of carapace gone; epizoa collected | Galveston | F | Yes/fish & worms | Liver, kidney, visceral fat | Heart exhibits pinhole on ventral surface of right atrium; Rt. anterior muscles exhibit severe hemorrhage |
| 90071104 (July 10-13) | CC | 52.2 | 2 | Scutes sloughing on shell; epizoa collected | Galveston | M | Yes/worms | Liver, kidney, visceral fat, blood, G.I. tract | Right 3rd Coastal scute exhibits two barnacle scars which open into the body cavity |
| 90071105 (July 10-13) | CC | 56.0 | 2 | Scutes sloughing on shell & head; carapace disarticulating; epizoa collected | Galveston | M | Yes/worms | Liver, kidney, visceral fat, blood | |
| 90071307 (July 10-13) | CC | 56.4 | 3 | Scutes sloughing on shell, head & flippers; carapace disarticulating | Matagorda | F | Yes/crabs, worms & fish | Liver, kidney | |
| 90072501 (Post-July 13) | CC | 62.5 | 2 | Scutes sloughing on shell; left rear flipper exhibits healed wound; epizoa collected | Brazoria | F | Yes/crabs & fish | Liver, kidney, visceral fat, blood | Left lung ruptured (surrounding tissue necrotic) |
| 90102001 (Post-July 13) | CC | 51.2 | 2 | Epizoa collected from carapace | Galveston | F | Yes/tissue, mollusks & worms | Liver, kidney, visceral fat, blood | |

* CC = loggerhead, *Caretta caretta*

** 2 = moderately decomposed; 3 = severely decomposed

Table 2. Heavy metal analysis ($\mu\text{g/g}$ or ppm wet weight) in 10 loggerhead sea turtles as related to the stranding event of 10-13 July 1990.

| TURTLE ID | | Pre-July 10 | | <- 10 - 13 July -> | | | | | | | Post-July 13 | |
|-----------|--------|-------------|----------|--------------------|----------|--------------------|--------------------|--------------------|----------|--------------------|--------------|--|
| | | 90030801 | 90051102 | 90071001 | 90071002 | 90071101 | 90071104 | 90071105 | 90071307 | 90072501 | 900102001 | |
| Zn | Liver | 33.84 | 36.76 | 18.61 (20.17) | 19.53 | 33.84 (24.87) | 24.88 (22.86) | 22.18 | 35.34 | 21.89 (15.06) | 19.22 | |
| Zn | Kidney | 61.54 | 25.25 | 28.05 | 24.55 | 30.84 | 28.40 | 40.87 (29.80) | 54.71 | 23.97 (25.15) | 19.64 | |
| Hg | Liver | 0.204 | 2.461 | 0.168 | 0.215 | 0.155 | 0.160 | 0.063 | 0.124 | 0.160 | 0.072 | |
| Hg | Kidney | 0.292 | 0.601 | 0.112 | 0.170 | 0.169 | 0.133 | 0.047 | 0.159 | 0.051 | 0.048 | |
| Fe | Liver | 614.78 | 443.82 | 168.52 (188.33) | 457.41 | 393.47 (317.31) | 298.56 (341.57) | 412.69 | 234.16 | 466.49 (401.46) | 413.69 | |
| Fe | Kidney | 317.50 | 515.13 | 257.14 | 158.44 | 211.62 | 266.71 | 418.52 (230.60) | 196.59 | 261.07 (182.56) | 295.09 | |
| As | Liver | 5.22 | 4.74 | 4.93 (4.66) | 4.51 | 4.81 (5.49) | 3.55 (4.11) | 4.56 | 8.29 | 2.06 (3.12) | 2.12 | |
| As | Kidney | 7.80 | 5.95 | 7.39 | 6.90 | 7.26 | 5.70 | 4.48 (4.25) | 7.86 | 4.20 (2.28) | 2.38 | |
| Cd | Liver | 7.29 | 3.06 | 3.39 | 1.09 | 4.53 | 2.75 | 5.37 | 4.61 | 3.49 | 1.68 | |
| Cd | Kidney | 47.36 | 5.87 | 16.15 | 3.63 | 20.47 | 12.93 | 26.19 | 39.82 | 9.39 | 7.03 | |
| Se | Liver | 4.62 | 5.13 | 2.92 (3.29) | 2.30 | 4.87 (4.01) | 3.76 (3.94) | 1.59 | 3.30 | 2.97 (3.31) | 2.50 | |
| Se | Kidney | 6.08 | 1.47 | 5.02 | 2.46 | 4.08 | 4.44 | 5.13 (3.96) | 3.29 | 4.50 (3.62) | 2.85 | |
| Pb | Liver | 0.05 | 0.68 | 0.03 | 0.05 | 0.06 | 0.02 | 0.07 | 0.05 | 0.03 | 0.05 | |
| Pb | Kidney | 0.25 | 0.34 | 0.17 | 0.17 | 0.14 | 0.12 | 0.14 | 0.15 | 0.05 | 0.12 | |
| Cu | Liver | 5.55 | 5.08 | 7.87 | 3.73 | 5.09 | 2.69 | 3.32 | 5.30 | 5.38 | 3.41 | |
| Cu | Kidney | 2.32 | 1.29 | 1.10 | 1.43 | 1.28 | 1.55 | 2.01 | 1.46 | 1.34 | 1.43 | |

() = Neutron activation data; other is atomic absorption

Table 3. Concentration (mean \pm S.D.) of metals in the kidney and liver of loggerhead turtles related to the stranding event of 10-13 July 1990 ($\mu\text{g/g}$ wet weight).

| Metal | Kidney | | | Liver | | |
|-------|--------------------|----------------------|---------------------|-------------------|----------------------|---------------------|
| | Control (4)* | Stranding Event (6)* | All (10)* | Control (4)* | Stranding Event (6)* | All (10)* |
| Zn | 31.85 \pm 19.87 | 34.57 \pm 11.31 | 33.78 \pm 14.08 | 27.93 \pm 8.66 | 25.73 \pm 7.22 | 26.61 \pm 7.43 |
| Hg | 0.248 \pm 0.261 | 0.132 \pm 0.047 | 0.178 \pm 0.166 | 0.72 \pm 1.16 | 0.147 \pm 0.05 | 0.378 \pm 0.733 |
| Fe | 334.7 \pm 124.23 | 251.50 \pm 91.02 | 284.78 \pm 107.67 | 484.7 \pm 89.38 | 327.46 \pm 112.53 | 390.36 \pm 127.63 |
| As | 5.08 \pm 2.32 | 6.6 \pm 1.26 | 5.99 \pm 1.81 | 3.53 \pm 1.68 | 5.21 \pm 1.87 | 4.48 \pm 1.75 |
| Cd | 17.41 \pm 20.01 | 14.86 \pm 12.36 | 18.88 \pm 14.83 | 3.88 \pm 2.4 | 3.62 \pm 1.55 | 3.73 \pm 1.81 |
| Se | 3.72 \pm 2.0 | 4.07 \pm 1.03 | 3.93 \pm 1.4 | 3.80 \pm 1.26 | 3.12 \pm 1.14 | 3.4 \pm 1.18 |
| Pb | 0.19 \pm 0.13 | 0.15 \pm 0.02 | 0.16 \pm 0.08 | 0.20 \pm 0.31 | 0.047 \pm 0.019 | 0.109 \pm 0.2 |
| Cu | 1.59 \pm 0.48 | 1.47 \pm 0.30 | 1.52 \pm 0.37 | 4.85 \pm 0.98 | 4.67 \pm 1.86 | 4.54 \pm 1.89 |

*() = Total number of animals

Table 4. Concentration of Metals Considered to be Normal in the Kidney, Liver, and Muscle of the Cow, Dog, Horse, and Pig; and Levels Reported in these organs when animals have died of metal poisoning (ug/g wet weight).*

| Animal | Metal | | | | | | |
|---------------|-----------|-----------|-----------|----------|------------|----------|----------|
| | As | Cd | Cu | Pb | Hg | Mn | Zn |
| Kidney | | | | | | | |
| Cow | | | | | | | |
| Normal | 0.018-0.4 | 0.05-1.5 | 4-6 | 30-150 | .008-0.09 | 0.2-2.0 | 10-25 |
| Toxic | 3.5-53 | 100-250 | 10-122 | ? | 4.3-200 | 5-700 | 100-480 |
| Dog | | | | | | | |
| Normal | <0.2 | 0.12-0.18 | 5-15 | 75-260 | | 0.1-2.5 | 16-30 |
| Toxic | >10.0 | 4-17 | >20 | ? | | 10-50 | 295 |
| Horse | | | | | | | |
| Normal | <0.4 | 0.05-10 | 7.3-9.3 | 35-150 | | 0.7-2.0 | |
| Toxic | >10.0 | 75-170 | 30-40 | 130-340 | | 5-200 | |
| Pig | | | | | | | |
| Normal | 0.003-0.1 | 0.15-0.99 | 7-10 | | <0.01-0.09 | <0.7 | 15-30 |
| Toxic | 10-70 | >270 | 10-1200 | | 10-200 | 1.4-2.9 | 190-36 |
| Liver | | | | | | | |
| Cattle | | | | | | | |
| Normal | 0.004-0.4 | 0.02-1.0 | 35-100 | 45-300 | .0007-0.06 | 0.1-1.0 | 25-100 |
| Toxic | 2.0-100 | 50-160 | 250-800 | ? | 2-40 | 5-300 | 120-500 |
| Dog | | | | | | | |
| Normal | <0.2 | 0.037 | 30-100 | 100-300 | | 0.1-3.5 | 30-70 |
| Toxic | >10.0 | 1-7 | 400-3000 | ? | | 50-200 | 369 |
| Horse | | | | | | | |
| Normal | 0.4 | 0.01-5 | 4.0-7.5 | 100-300 | | 0.08-1.4 | |
| Toxic | 7-15 | 80+ | 1000-1500 | 600-1000 | | 4-500 | |
| Pig | | | | | | | |
| Normal | 0.003-.2 | 0.04-0.5 | 5-25 | 100-200 | <0.01-0.03 | <0.7 | 40-90 |
| Toxic | 6.3-28 | 13+ | 150-15000 | 400+ | 5-150 | ? | 500-3100 |
| Muscle | | | | | | | |
| Cattle | | | | | | | |
| Normal | .004-.024 | 1.2-1.5 | 45-54 | | 0.07-0.15 | 2.01 | |
| Toxic | | | | | | | |
| Dog | | | | | | | |
| Normal | | | | | 0.5-1.5 | | |
| Toxic | | | | | | | |
| Horse | | | | | | | |
| Normal | <0.01 | | | | | | |
| Toxic | | | | | | | |
| Pig | | | | | | | |
| Normal | | | | | 0.06-0.32 | | |
| Toxic | | | | | | | |

* Robert Puls, 1988

Table 5. Metal concentrations in field-collected fresh water and sea turtles.

| Metal | Species | Location | Concentration (µg/g wet weight) | | Tissue | Number of Animals | Sex | Source |
|----------------------------|-------------------------------|--------------------------------|------------------------------------|-------------|---------|-------------------------|---------------------------------|--|
| | | | | | | | | |
| Zinc | <i>Caretta caretta</i> | Seashores, Ga, SC | 2.08 | egg yolks | albumin | unknown | - | Hillestad <i>et al.</i> (1974) |
| | | | 6.0 | | | unknown | - | |
| | <i>Caretta caretta</i> | National Seashores, FL, GA, NC | 25.6 - 28.0 | egg yolks | | 96 | - | Stoneburner <i>et al.</i> (1980) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #1, NJ | 9.93 | kidney | | 8 | M | Albers <i>et al.</i> (1986) |
| | | | 50.4 | liver | | 8 | M | |
| | <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | 9.79 | kidney | | 3 | F | Albers <i>et al.</i> (1986) |
| | | | 39.0 | liver | | 3 | F | |
| | | | 10.5 | kidney | | 8 | M | |
| | | | 30.7 | liver | | 8 | M | |
| | <i>Chelydra serpentina</i> | Reference wetland, MD | 8.80 | kidney | | 7 | M | Albers <i>et al.</i> (1986) |
| | | 27.7 | liver | | 7 | M | | |
| | | 9.60 | kidney | | 6 | F | | |
| <i>Chelydra serpentina</i> | North AL reservoir | 29.3 | liver | | 6 | F | | |
| | | 36.8 | muscle | | 28 | M&F | Dycus (1986) | |
| | <i>Caretta caretta</i> | Texas Gulf | 33.78 | kidney | | 28 | M&F | |
| | | | 26.61 | liver | | 10 | M&F | Sis <i>et al.</i> (1992 - this report) |
| Mercury | <i>Kinosternon flavescens</i> | Rice field, TX | 0.12 | whole body | | 3 | unknown | Flickinger and King (1972) |
| | <i>Trechmanys scripta</i> | Rice field, TX | 0.08 | whole egg | | 2 | unknown | Flickinger and King (1972) |
| | <i>not reported</i> | Little Piney River | n.d. | unlaid eggs | | 4 | | |
| | <i>Pseudemys scripta</i> | Elephant Butte Reservoir, NM | 0.02 - 0.04 | unknown | | 5 | | Bertrand (1974) |
| | | | 0.78 | kidney | | 8 | M&F | Kidd <i>et al.</i> (1974) |
| | <i>Trionyx spinifer</i> | Elephant Butte Reservoir, NM | 0.79 | liver | | 15 | M&F | |
| | | | 0.84 | kidney | | 11 | M&F | Kidd <i>et al.</i> (1974) |
| | | | 0.84 | liver | | 15 | M&F | |
| | <i>Caretta caretta</i> | National seashores, FL, GA, NC | 0.14 - 0.48 | egg yolks | | 96 | | Stoneburner <i>et al.</i> (1980) |
| | <i>Chelydra serpentina</i> | 3 river & 1 lake sites, NM | 0.02 | fat | | M | 9 | Helwig and Hora (1983) |
| | | 0.12 | muscle | | M | 10 | | |
| | | 0.03 | fat | | F | 5 | | |
| | | 0.21 | muscle | | F | 5 | | |
| | | 0.03 | fat | | unknown | 1 | | |
| | | 0.1 | muscle | | unknown | 2 | | |
| <i>Chelydra serpentina</i> | Contaminated wetland #1, NJ | 0.55 | kidney | | M | 8 | Albers <i>et al.</i> (1986) | |
| | | 1.28 | liver | | M | 8 | | |
| <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | 0.41 | kidney | | F | 3 | Albers <i>et al.</i> (1986) | |
| | | 1.27 | liver | | F | 3 | | |
| | | 0.39 | kidney | | M | 8 | | |
| | | 0.60 | liver | | M | 8 | | |
| <i>Chelydra serpentina</i> | North AL Reservoir | 1.85 | liver | | - | 28 | Dycus (1986) | |
| | | .23 | muscle | | - | 15 | | |
| <i>Chelydra serpentina</i> | Tennessee Reservoir | .27 | liver | | - | 6 | Dycus and Lowery (1986) | |
| <i>Chelydra serpentina</i> | White Oak Lake, TN | 1.3 | kidney | | - | 12 | Meyers-Schone and Walton (1990) | |
| | | 34 | muscle | | - | 12 | | |
| <i>Chelydra serpentina</i> | Beardon Creek, TN | .17 | kidney | | - | 9 | Meyers-Schone and Walton (1990) | |
| | | .10 | muscle | | - | 9 | | |
| <i>Pseudemys scripta</i> | White Oak Lake, TN | 0.64 | kidney | | - | 12 | Meyers-Schone and Walton (1990) | |
| | | .10 | muscle | | - | 12 | | |

Table 5. (Continued)

| Metal | Species | Location | Concentration (µg/g wet weight) | Tissue | Number of Animals | Sex | Source |
|-----------------|----------------------------|---|--|---|---------------------------------|---------------------------------|--|
| Mercury (cont.) | <i>Pseudemys scripta</i> | Beardon Creek, TN | | kidney muscle | | - | Meyers-Schone and Walton (1990) |
| Iron | <i>Caretta caretta</i> | National Seashores, FL, GA, NC | | egg yolks | 96 | - | Stoneburner <i>et al.</i> (1980) |
| | <i>Caretta caretta</i> | Texas Gulf | 24.8 - 26.0 284.78 390.36 | kidney liver | 10 10 | M&F M&F | Sis <i>et al.</i> (1992 - this report) |
| Arsenic | <i>Caretta caretta</i> | Texas Gulf | | kidney | 10 | M&F | Sis <i>et al.</i> (1992 - this report) |
| Cadmium | <i>Caretta caretta</i> | Seashores, GA, AL | | egg yolks albumin | unknown unknown | - | Hillestad <i>et al.</i> (1974) |
| | <i>Trionyx spiniferus</i> | River that received effluent from plating industries, TN | | kidney | 12 | F | Robinson and Wells (1975) |
| | <i>Caretta caretta</i> | National Seashores, FL, GA, NC | 0.01 - 0.07 | egg yolks | 96 | - | Stoneburner <i>et al.</i> (1980) |
| | <i>Chelydra serpentina</i> | 5 river & 1 lake site, NM | 0.010 0.012 | muscle muscle | 8 4 | M F | Helwig and Hora (1983) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #1, NJ | 0.25 0.10 0.10 0.08 | kidney liver kidney liver | 8 8 3 3 | M M F F | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | 0.09 0.08 | kidney liver | 8 8 | M M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Reference wetland, MD | 0.07 0.07 0.07 | kidney liver kidney | 7 7 6 | M M F | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Tennessee Reservoir | 0.024 0.03 0.33 | fat liver liver | 6 6 28 | - - - | Dycus and Lowery (1986) |
| | <i>Chelydra serpentina</i> | North AL Reservoirs | 0.28 | muscle | 28 | - | Dycus (1986) |
| | <i>Caretta caretta</i> | Texas Gulf | 18.88 3.73 | kidney liver | 10 10 | M&F M&F | Sis <i>et al.</i> (1992 - this report) |
| Selenium | <i>Chelydra serpentina</i> | Tennessee Reservoir | 0.69 0.15 | liver fat | 6 6 | - - | Dycus and Lowery (1986) |
| | <i>Caretta caretta</i> | Texas Gulf | 3.93 3.4 | kidney liver | 10 10 | - - | Sis <i>et al.</i> (1992 - this report) |
| Lead | <i>Caretta caretta</i> | Seashores, GA, SC | | egg yolks albumin | unknown unknown | - | Hillestad <i>et al.</i> (1974) |
| | <i>Caretta caretta</i> | National Seashores, FL, GA, NC | 1.39 - 0.76 | egg yolks | 96 | - | Stoneburner <i>et al.</i> (1980) |
| | <i>Terrapene carolina</i> | Woodland area near Pb smelter, MD | 51.8 65.5 21.6 24.3 6.00 0.35 0.20 | humans femur liver kidney blood skin lung | 4 4 4 4 4 4 4 | M M M M M M M | Beresford <i>et al.</i> (1981) |

Table 5. (Continued)

| Metal | Species | Location | Concentration ($\mu\text{g/g}$ wet weight) | Tissue | Number of Animals | Sex | Source |
|-----------------|----------------------------|--------------------------------|--|-----------|-------------------------|-----|--|
| Lead (cont.) | <i>Terrapene carolina</i> | Reference woodland, WV | 4.51 | humerus | 1 | M | Beresford <i>et al.</i> (1981) |
| | | | 5.55 | femur | 1 | M | |
| | | | 2.21 | liver | 1 | M | |
| | | | 4.83 | kidney | 1 | M | |
| | | | 0.22 | blood | 1 | M | |
| | | | n.d. | skin | 1 | M | |
| | | | n.d. | lung | 1 | M | |
| | | | 3.41 | humerus | 1 | M | |
| | | | 3.21 | femur | 1 | F | |
| | | | 0.80 | liver | 1 | F | |
| | | | 0.77 | kidney | 1 | F | |
| | | | n.d. - 0.15 | blood | 1 | F | |
| | | | n.d. - 0.16 | skin | 1 | F | |
| | | | n.d. | lung | 1 | F | |
| Copper | <i>Chelydra serpentina</i> | Contaminated wetland #1, NJ | 0.19 | kidney | 8 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | n.d. | kidney | 8 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Reference wetland, MD | n.d. | kidney | 3 | F | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | | n.d. | liver | 3 | F | |
| | <i>Chelydra serpentina</i> | | 0.10 | kidney | 8 | M | |
| | <i>Chelydra serpentina</i> | | 0.12 | liver | 8 | M | |
| | <i>Chelydra serpentina</i> | Tennessee Reservoir | 0.07 | kidney | 7 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | | 0.07 | liver | 7 | M | |
| | <i>Chelydra serpentina</i> | | 0.16 | kidney | 6 | F | |
| | <i>Chelydra serpentina</i> | | n.d. | liver | 6 | F | |
| | <i>Caretta caretta</i> | Texas Gulf | .65 | liver | 6 | - | Dycus and Lowery (1986) |
| | <i>Caretta caretta</i> | Texas Gulf | .26 | fat | 6 | - | Dycus and Lowery (1986) |
| | <i>Caretta caretta</i> | Seashores, GA, SC | 0.16 | kidney | 10 | M&F | Sis <i>et al.</i> (1992 - this report) |
| | <i>Caretta caretta</i> | | 0.109 | liver | 10 | M&F | |
| | <i>Caretta caretta</i> | | 2.08 | egg yolks | unknown | - | |
| | <i>Caretta caretta</i> | | 6.0 | albumin | unknown | - | Hillestad <i>et al.</i> (1974) |
| | <i>Chelydra serpentina</i> | National Seashores, FL, GA, NC | 1.73 - 2.30 | egg yolks | 96 | - | Stoneburner <i>et al.</i> (1980) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #1, NJ | 1.01 | kidney | 8 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | 9.72 | liver | 8 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | | 1.73 | kidney | 3 | F | |
| | <i>Chelydra serpentina</i> | | 5.17 | liver | 3 | F | |
| | <i>Chelydra serpentina</i> | | 1.73 | kidney | 8 | M | |
| | <i>Chelydra serpentina</i> | Reference wetland, MD | 2.08 | liver | 8 | M | Albers <i>et al.</i> (1986) |
| | <i>Chelydra serpentina</i> | Tennessee Reservoir | 0.82 | kidney | 7 | M | Dycus and Lowery (1986) |
| | <i>Chelydra serpentina</i> | | 1.28 | liver | 7 | M | |
| | <i>Chelydra serpentina</i> | | 1.07 | kidney | 6 | F | |
| | <i>Chelydra serpentina</i> | | 1.57 | liver | 6 | F | |
| Caretta caretta | <i>Chelydra serpentina</i> | Contaminated wetland #2, NJ | 1.9 | liver | 6 | - | Dycus and Lowery (1986) |
| | <i>Chelydra serpentina</i> | North AL Reservoir | 2.2 | liver | 28 | - | Dycus (1986) |
| | <i>Chelydra serpentina</i> | Texas Gulf | .96 | muscle | 28 | - | Dycus (1986) |
| | <i>Caretta caretta</i> | Texas Gulf | 1.52 | kidney | 10 | M&F | Sis <i>et al.</i> (1992 - this report) |
| | | | 4.54 | liver | 10 | M&F | Sis <i>et al.</i> (1992 - this report) |

Table 6. Summary of necropsy data for 10 sea turtles utilized for organochlorine and PAHs analyses related to the stranding event of 10-13 July 1990.

| ID AND (TIME PERIOD) | SPECIES* | SCL(cm) STRAIGHT CARAPACE LENGTH (cm) | CONDITION CODE** | EXTERNAL ABNORMALITIES | COUNTY | SEX | G.I. CONTENTS COLLECTED/ MAJOR ITEMS | TOXICOLOGICAL SAMPLES | MISCELLANEOUS COMMENTS |
|----------------------------|----------|--|---------------------|---|-----------|-----|--|---|--|
| 90031201 (Pre-July 10) | LK | 61.5 | 2 | Scutes sloughing on carapace & plastron | | F | Yes/crabs | Liver, kidney, visceral fat, blood | |
| 90042501 (Pre-July 10) | LK | 64.2 | 2 | Skin sloughing on head & all flippers | Galveston | F | Yes/crabs | Liver, kidney, visceral fat, blood | Heart exhibits small hole in dorsal surface of ventricle |
| 90071001 (July 10-13) | CC | 57.2 | 2 | Black material (tar) collected from plastron & Lt. rear flipper; epizoa collected | Galveston | M | Yes/ crabs, worms & anemones | Liver, kidney, visceral fat, blood | Dorsal tip of Rt. front flipper exhibits healed wound |
| 90071002 (July 10-13) | CC | 76.5 | 2 | Skin sloughing on head; epizoa collected | Galveston | F | Yes/worms | Liver, kidney, visceral fat, blood | Fish hook & fishing line found in tissue dorsal to esophagus; right atrium of heart exhibits air bubble |
| 90071101 (July 10-13) | CC | 56.8 | 2 | Scutes sloughing on shell & flippers; left marginal bones of carapace gone; epizoa collected | Galveston | F | Yes/fish & worms | Liver, kidney, visceral fat | Heart exhibits pinhole on ventral surface of right atrium; Rt. anterior muscles exhibit severe hemorrhage |
| 90071104 (July 10-13) | CC | 52.2 | 2 | Scutes sloughing on shell; epizoa collected | Galveston | M | Yes/worms | Liver, kidney, visceral fat, blood, G.I. tract | Right 3rd Coastal scute exhibits two barnacle scars which open into the body cavity |
| 90071105 (July 10-13) | CC | 56.0 | 2 | Scutes sloughing on shell & head; carapace disarticulating; epizoa collected | Galveston | M | Yes/worms | Liver, kidney, visceral fat, blood | |
| 90071306 (July 10-13) | LK | N/A | 3 | Scutes sloughing on shell, head & flippers; shell disarticulating | Matagorda | N/A | Yes/crabs | Liver, kidney, visceral fat | |
| 90072501 (Post-July 13) | CC | 62.5 | 2 | Scutes sloughing on shell; left rear flipper exhibits healed wound; epizoa collected | Brazoria | F | Yes/crabs & fish | Liver, kidney, visceral fat, blood | Left lung ruptured (surrounding tissue necrotic) |
| 90102001 (Post-July 13) | CC | 51.2 | 2 | Epizoa collected from carapace | Galveston | F | Yes/tissue, mollusks & worms | Liver, kidney, visceral fat, blood | |

* LK = Kemp's ridley, *Lepidochelys kempi*; CC = loggerhead, *Caretta caretta*

** 2 = moderately decomposed

Table 8. Organochlorine pesticides and polyaromatic hydrocarbons analyses of sea turtles related to the stranding event of 10-13 July 1990.

| TURTLE ID | Pre-July 10 | | <- July 10 - 13 -> | | | | | | Post-July 13 | |
|---------------------------------|-------------|---------|--------------------|---------|---------|---------|---------|----------|--------------|----------|
| | LK31201 | LK42501 | CC71001 | CC71002 | CC71101 | CC71104 | CC71105 | LK71306 | CC72501 | CC102001 |
| Lipid wt. | 1.89 | 0.75 | 1.72 | 1.50 | 0.81 | 0.39 | 0.45 | 0.20 | 1.90 | 0.29 |
| Tissue wt. | 3.00 | 3.02 | 2.92 | 4.09 | 2.29 | 2.40 | 2.42 | 3.26 | 2.24 | 2.37 |
| HEPTACHLOR total ng | 2984.38 | N.D. | 873.85 | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |
| Total ng/g lipid | 1579.04 | | 508.05 | | | | | | | |
| Total ng/g tissue | 994.79 | | 299.26 | | | | | | | |
| SUM DDT total ng* | | | | | | | | | | |
| Total ng/g lipid | N.D. | N.D. | 1164.80 | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |
| Total ng/g tissue | | | 677.21 | | | | | | | |
| | | | 398.80 | | | | | | | |
| DIELDRIN total ng | N.D. | N.D. | 555.22 | 718.35 | N.D. | N.D. | N.D. | 933.66 | N.D. | N.D. |
| Total ng/g lipid | | | 322.80 | 478.90 | | | | 4668.28 | | |
| Total ng/g tissue | | | 190.14 | 175.64 | | | | 286.40 | | |
| ENDRIN ALDEHYDE total ng | N.D. | N.D. | 2493.52 | N.D. | N.D. | N.D. | N.D. | 2659.22 | N.D. | N.D. |
| Total ng/g lipid | | | 1449.72 | | | | | 13296.11 | | |
| Total ng/g tissue | | | 853.95 | | | | | 815.71 | | |
| beta-BHC total ng | 71010.3 | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |
| Total ng/g lipid | 37571.6 | | | | | | | | | |
| Total ng/g tissue | 23670.1 | | | | | | | | | |
| Methoxychlor | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |
| Total ng/g lipid | | | | | | | | | | |
| Total ng/g tissue | | | | | | | | | | |
| Heptachlor epoxide | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | 192.30 | N.D. |
| Endrin | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | 101.21 | N.D. |
| alpha-BHC | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | 85.85 | N.D. |
| Endosulfan S04 | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | | N.D. |
| Kepone | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | | N.D. |
| 2,4-dichlorophenoxy acetic acid | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |

N.D. = not detected

Total ng/g = parts per billion

* SUM DDT = pp'-DDE, pp'-DDD and pp'-DDT

Table 10. Concentration of metals in the liver of whales and other marine mammals ($\mu\text{g/g}$ wet weight).

| Common Name Genus, Species | Metal | | | | | | | | Reference |
|---|---------------------------------------|---|--------------------------------|---------------------------------------|--|--------------------------------|---|-----------------------------------|--|
| | As | Cd | Cu | Fe | Hg | Pb | Se | Zn | |
| Bowhead Whale (<i>Balaena mysticetus</i>) 1980 (2)** 1986 (6) 1988 (4) | 0.024-0.085 0.02-0.27 0.26-0.69 | 1.5 3.98-8.32 8.65-13.69 11.3-19.8 | 3.09 5.27-9.44 5.32-9.07 | N/M*** 46.09-108.52 86.1-138.05 | 0.009 0.007-0.008 0.007-0.008 19.2-45.4 | 0.12 0.01-0.07 0.03-0.05 | 0.05-0.11 1.0-1.75 1.06-1.43 32.9-61.6 | 43.6 6.12-12.741 10.22-13.2 | Byrne <i>et al.</i> (1985) ours ours |
| Short Finned Pilot Whale (17) (<i>Globicephala macrorhyncha</i>) | | | | | | | | | Gaskin <i>et al.</i> (1974); Stoneburner (1978) |
| Long Finned Pilot Whale (220) (<i>Globicephala melaleuca</i>) | 0.01-2.91 | 0.01-12.5 | 2.39-5.8 | 557 | 0.08-150 | 0.02-0.447 | 0.03-44 | 18.84-198.33 | Anderson <i>et al.</i> (1987); Denton <i>et al.</i> (1980); Jean-Courvot <i>et al.</i> (1989); Muir <i>et al.</i> (1988) |
| Beluga Whale (12) (<i>Delphinapterus leucas</i>) | 0.22-1.86 | 0.90-4.3 | 20.4 | | 2.45-56.3 | 0.2-1.8 | | 26.2-33.6 | Blight and Armstrong (1971); Harms <i>et al.</i> (1978); Wagemann and Muir (1984); Sergeant (1980); Huschenbeth (1977) |
| Botlenose Whale (1) (<i>Hyperoodon amipullatus</i>) | | 5.6 | 2.8 | | 0.38 | 0.18 | | 23 | Harms <i>et al.</i> (1978) |
| Goosebeak Whale (4) (<i>Ziphius cavirostris</i>) | | 37-64 | 3.2-7.1 | 472-528 | | <0.5 | | 40.59 | Knapp and Jickells (1983) |
| Narwhale (69) (<i>Monodon monoceros</i>) | | 0.28-130.8 | 1.98-20.31 | | 0.57-13.1 | 0.01-0.57 | 0.62-14.8 | 15.5-87.9 | Wagemann and Muir (1984) |
| Mink Whale (148) (<i>Balaenoptera acutorostrata</i>) | | 2.16-32.72 | 2.27-8.31 | 0.14-7.9 | 0.02-0.41 | 0.03-0.63 | | 24.26-57.07 | Wagemann <i>et al.</i> (1983) |
| Pigmy Sperm Whale (1) (<i>Physeter macrocephalus</i>) | | 0.22-7.60 | 2.1-14.44 | | | 1.5 | 10.09-15.04 | | Honda <i>et al.</i> (1987) |
| Botlenose Dolphin (5) (<i>Tursiops truncatus</i>) | | 0.04 | 3.0-4.4 | | 5.18 | 0.4 | | | Johansen <i>et al.</i> (1980) |
| White Beaked Dolphin () (<i>Lagenorhynchus albirostris</i>) | | 0.05-2.33 | 0.97-8.89 | | 0.13-1.61 | 0.008-0.34 | 1.0-3.43 | 21-30 | Odell and Asper (1976) |
| Dall's Porpoise (3) (<i>Phocoenoides dalli</i>) | | 0.006-20.6 | 5.39-82.2 | 285-408 | 6.38 | <0.02 | | | Arima and Nagakura (1979) |
| Common Harbor Porpoise (27) (<i>Phocoena phocoena</i>) | 0.03-1.9 | 0.007-1.20 | 2.6-15 | | 0.28-192 | 0.06-5.3 | 0.2-79 | 12.05-37.67 | Windom (1972) Muir <i>et al.</i> (1988) |
| Striped Dolphin (120) (<i>Stenella caeruleoalba</i>) | | 0.04-11.1 | 3.57-15.2 | 55.8-200+ | 1.70-485 | 0.03-0.64 | 1.07-48.6 | 27.5-46.14 | Fujise <i>et al.</i> (1988) |
| Long Snout Dolphin () (<i>Stenella longirostris</i>) | | | | | 6-13 | | | | Anderson & Rebsdorff (1976); Harms <i>et al.</i> (1978); Falconer <i>et al.</i> (1983); Gaskin <i>et al.</i> (1972, 1979); Huschenbeth (1977); Koeman <i>et al.</i> (1972) |
| Swainson Dolphin () (XXXXXX xxxxxx) | 0.15-0.19 | 0.02-0.06 | | | 0.37-11.0 | | 0.6-3.20 | 26.5-109 | Fujise <i>et al.</i> (1988); Honda <i>et al.</i> (1982, 1983); Iliano <i>et al.</i> (1984a, b) |
| Spotted Dolphin () (<i>Stenella attenuata</i>) | | 8.7 | 5.4 | | | | | | Gaskin <i>et al.</i> (1974) |
| | | | | | | | | 59-66 | Koeman <i>et al.</i> (1972) |
| | | | | | 6.2 | | | 33.50 | Andre (1988) |

** () = Total number animals reported where it was possible to determine numbers

*** N/M = Not measured

APPENDIX A

TURTLE QC

| TISSUE | | Zn ppm | Hg ppm | Fe ppm | As ppm | Cd ppm | Se ppm | Pb ppm | Cu ppm |
|--------|----------------|------------|-------------|------------|------------|---------------|-------------|-------------|-------------|
| DOLT | Mean | 95.20 | .25553 | 636.76 | 7.17 | 4.57 | 7.39 | 1.01 | 15.01 |
| | Std. deviation | 14.1 | .0430 | 77.4 | 1.1 | 0.8 | 0.8 | 0.1 | 2.1 |
| | REPORTED | 92.5 ± 2.3 | 255 ± 0.037 | 712 ± 48 | 10.1 ± 1.4 | 4.18 ± 0.28 | 7.34 ± 0.42 | 1.36 ± 0.29 | 20.8 ± 1.2 |
| | | | | | | | | | |
| | | | | | | | | | |
| DORM | Mean | 17.44 | .74737 | 69.34 | 13.97 | 0.57 | 1.59 | 0.38 | 4.24 |
| | Std. deviation | 0.20 | | 9.01 | 0.97 | 0.69 | 0.39 | 0.18 | 0.38 |
| | REPORTED | 21.3 ± 1 | .798 ± .074 | 63.6 ± 5.3 | 17.7 ± 2.1 | 0.086 ± 0.012 | 1.62 ± 0.12 | 0.4 ± 0.12 | 5.22 ± 0.33 |
| | | | | | | | | | |